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Enhanced production of a recombinant thermostable GH family 12 endo-1,4-β-glucanase from *Thermotoga petrophila* in a mesophilic expression host through various cultivation and induction strategies

Irfana Mariam[1], **Ikram ul Haq**[2] and **Fatima Akram**[2]
[1]Queen Mary College, Pakistan
[2]Government College University, Pakistan

Enhancing heterologous expression and production of thermostable cloned proteins in mesophilic expression host system is the most prominent aspect of revolutionized biochemistry. Inadvertent induction and unintentionally preparation of cultivation media give deprived expression of recombinant enzymes in engineered cells. Therefore, to augment the expression and production of an industrially pertinent endo-1,4-β-glucanase through various cultivation and induction strategies, propagated in *Escherichia coli* BL21 CodonPlus (DE3)-RIPL mesophilic host. After exploration of various modifying cultivation media and induction parameters, it has demonstrated that high-cell-density or dry cell weight (DCW) and optimal expression of endo-1,4-β-glucanase were obtained in 3×ZYBM9 medium after 72 h inducement at 22°C, induced the culture either with 0.5 mM IPTG/100 mM lactose after heat shock treatment (42°C for 60 minutes) when OD 600 nm reached at 0.6. Recombinant extracellular soluble enzyme activity was improved by 7.78 and 6.18-fold in 3×ZYBM9 and ZYBM9, respectively under optimal cultivation conditions. Using M9NG and YNG auto-induction medium, activity was 6.5 and 4.76-fold increased after 72 h incubation at 22°C with agitation (200 rev min-1). Similar results were obtained after studying the fermentation process at the bioreactor level using optimal parameters. Hence, the results showed that the effective process strategy is essential to enhance engineered cell mass (production) and enzyme expression from small to large-scale. Highly thermostable endo-1,4-β-glucanase cloned from *Thermotoga petrophila*, is the most interesting and suitable candidate for numerous industrial applications.

majied.irfana@yahoo.com